Supporting Information for

Towards an Artificial Aldolase

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Preparation and characterization of Polystyrene resin **2** (PS-PEG derived resin):

a) Reaction of PS-PEG-Br with sodium azide.

Sodium azide (0.114 g, 51 mmol) was added to a mixture of 1.4 g of NovaBioSyn™ resin (Br-PEG-PS, $f = 0.25$ mmol/g) in 10 mL of DMSO, and heated at 60 °C for 18 h. After cooling, the suspension was filtered off and the resin was washed with H₂O, THF, THF-MeOH, MeOH and THF. The solid was dried *in vacuo* for 24 h at 40 °C

IR (ATR): 3045, 2865, 2100, 1638, 1452 cm⁻¹

Elemental analysis (%): C, 64.95; H, 9.62; N, 1.06.

$f = 0.25$ mmol/g

b) Cycloaddition of *tert*-butyl (2*S*,4*R*)-*N*-Boc-4-propargyloxyprolinate with N₃-PEG-PS.

tert-Butyl (2*S*,4*R*)-*N*-Boc-4-propargyloxyprolinate (0.46 g, 1.2 mmol), [SI] *N,N*-diisopropylethylamine (0.60 mL, 4.2 mmol) and copper iodide (0.005 g, 0.003 mmol) were added to a suspension of N₃-PEG-PS (1 g, *f* = 0.25 mmol/g) in DMF:THF 1:1 at 35°C and shaken for 24 h. When the IR-signal of the azido group had completely disappeared, the reaction mixture was filtered off and the resin was washed with THF (250 mL), water (250 mL), THF (250 mL), THF-MeOH 1:1 (250 mL), MeOH (250 mL) and THF (250 mL). The solid was dried *in vacuo* for 24 hours at 40 °C.

IR (ATR): 3045, 2864, 1736, 1699, 1452, 1398 cm⁻¹

Elemental analysis (%): C, 64.94; H, 9.69; N, 1.10.

f = 0.19 mmol/g

c) Deprotection .

The doubly protected resin (1 g, $f = 0.19$ mmol/g) was swollen in CH_2Cl_2 (5 mL). After 10 minutes, trifluoroacetic acid (10 mL) was added and the reaction mixture was shaken at room temperature. When the IR-signals of the *t*Bu and carbonyl moieties in the protecting groups had completely disappeared, the reaction mixture was filtered off, and the polymer-supported proline **2** was washed with THF (containing 2% of Et_3N , 250 mL), water (250 mL), THF (250 mL), THF-MeOH 1:1 (250 mL), MeOH (250 mL) and THF (250 mL). The solid was dried *in vacuo* for 24 h at 40 °C.

IR (ATR): 3504, 3042, 2904, 2871, 1639, 1452, 1348 cm^{-1}

Elemental analysis (%): C, 64.86; H 9.10; N, 0.98.

$f = 0.17$ mmol/g

Preparation and characterization of Polystyrene resin **3** (ArgoPore™ derived resin):

a) Synthesis of (azidobenzyl)polystyrene.

Sodium azide (1.07 g, 16.5 mmol) was added to a mixture of 3 g of (chlorobenzyl)polystyrene (ArgoPore™, $f = 0.6\sim 1.1$ mmol/g) in 30 mL of DMSO. The mixture was heated for 18 h at 60°C. After cooling, the suspension was filtered off and the solid washed with H₂O, THF, THF-MeOH, MeOH and THF. The solid was dried *in vacuo* for 24 h at 40 °C.

IR (ATR): 3042, 2921, 2096, 1492, 1451 cm⁻¹

Elemental analysis (%): C, 86.05; H, 8.14; N, 4.44.

$f = 0.79$ mmol azido/g

b) Cycloaddition of *tert*-butyl (2*S*,4*R*)-*N*-Boc-4-propargyloxyprolinate with (azidobenzyl)polystyrene resin.

(2*S*,4*R*)-*N*-Boc-4-propargyloxy proline (0.46 g, 1.2 mmol), [*S*] *N,N*-diisopropylethylamine (0.60 mL, 4.2 mmol) and copper iodide (0.005g, 0.003 mmol) were added to a suspension of (azidobenzyl)polystyrene (1 g, *f* = 0.79 mmol/g) and DMF:THF 1:1 at 35°C and shaken for 24 h. When the IR-signal of the azido group had completely disappeared, the reaction mixture was filtered off and the resin was washed with THF (250 mL), water (250 mL), THF (250 mL), THF-MeOH 1:1 (250 mL), MeOH (250 mL) and THF (250 mL). The solid was dried *in vacuo* for 24 hours at 40 °C.

IR(ATR): 3041, 2922, 1698, 1686, 1600, 1450, 1400, 1366 cm^{-1}

Elemental analysis (%): N, 4.55; C, 81.88; H, 9.96.

f = 0.81 mmol/g

c) Deprotection.

The protected proline-functionalized resin obtained in the previous step (1 g, *f* = 0.81 mmol/g) was swollen in CH_2Cl_2 (5 mL). After 10 minutes, trifluoroacetic acid was added (10 mL) and the reaction mixture was shaken at r.t. When the IR-signals of *t*Bu and

carbonyl groups had completely disappeared, the reaction mixture was filtered off, and polymer-supported proline **3** was washed with THF (containing 2% of Et₃N, 250 mL), water (250 mL), THF (250 mL), THF-MeOH 1:1 (250 mL), MeOH (250 mL) and THF (250 mL). The solid was dried *in vacuo* for 24 hours at 40 °C.

IR(ATR): 3450, 2921, 1672, 1600, 1492, 1450 cm⁻¹

Elemental analysis (%): N, 4.54; C, 79.55; H, 7.09.

f = 0.81 mmol/g

Preparation and characterization of Polystyrene resin 4:

a) Preparation of propargyloxymethyl polystyrene

In a flame-dried flask, propargyl alcohol (0.237 mL, 4 mmol) was added dropwise to a suspension of sodium hydride in DMF (5 mL) at -20°C , and was stirred for 20 min at this temperature. Then, the reaction mixture was heated at room temperature, and transferred *via canula* to a suspension of Merrifield resin (3 g, 1 % DVB, $f = 0.8\sim 1$ mmol Cl/g) in THF. The reaction mixture was shaken at room temperature for 12 h. Then, MeOH (5 mL) was added, the reaction mixture was filtered off, and the resin washed sequentially with THF, H_2O , THF, THF-MeOH 1:1, MeOH and THF. The solid was dried *in vacuo* for 24 h at 40°C .

IR (ATR): 3298, 3059, 3025, 2918, 2848, 1601, 1492, 1451 cm^{-1} .

b) Cycloaddition of *tert*-butyl (2*S*,4*R*)-*N*-Boc-4-azidoprolinate with propargyloxymethyl polystyrene.

tert-Butyl (2*S*,4*R*)-*N*-Boc-4-azidoprolinate (Prepared according to literature methods, [S2]) (500 mg, 1.60 mmol), *N,N*-diisopropylethylamine (2.0 ml, 13.8 mmol) and copper iodide (5 mg, 0.026 mmol) were added to a suspension of propargyloxymethyl polystyrene (1.33 g, 1.06 mmol propargyl groups) in DMF:THF 1:1. The mixture was stirred at 35°C for 24 h, the reaction progress being monitored by FTIR. When the IR bands of the C≡CH groups had completely disappeared, the reaction mixture was filtered off, and the resin was sequentially washed with THF (250 mL), water (250 mL), THF (250 mL), THF-MeOH 1:1 (250 mL), MeOH (250 mL) and THF (250 mL). The solid was dried *in vacuo* for 24 hours at 40 °C.

IR(ATR): 3025, 2974, 2922, 2851, 1737, 1701, 1393, 1366, 1061, 1492, 1453 cm⁻¹

c) Deprotection.

Proline protected resin (1 g) was swollen in CH_2Cl_2 (5 mL). After 10 minutes, trifluoroacetic acid was added (10 mL), and the reaction mixture was shaken at room temperature. When the IR bands of the *t*Bu and carbonyl moieties in the protecting groups had completely disappeared, the reaction mixture was filtered off, and the resin was sequentially washed with THF (with 2% of Et_3N , 250 mL), water (250 mL), THF (250 mL), THF-MeOH 1:1 (250 mL), MeOH (250 mL) and THF (250 mL). The solid was dried *in vacuo* for 24 h at 40 °C.

IR(ATR): 3450, 3048, 2904, 1628, 1600, 1492, 1451 cm^{-1}

Elemental analysis (%): N, 3.16; C, 84.17; H, 8.30.

$f = 0.56 \text{ mmol/g}$

General procedure for the asymmetric aldol reaction in water catalyzed by resin **4**:

Polystyrene-supported proline **4** (10% mol) was added to a suspension of aldehyde (0.4 mmol) and cyclohexanone (2.0 mmol) in water (0.14 mL). The reaction mixture was stirred at r.t. for 24 hours. Then 2 mL of ethyl acetate were added, the mixture was filtered off, the resin was washed with ethyl acetate (4 x 2 mL) and the combined liquid phases were concentrated *in vacuo*. Purification by silica gel column chromatography (Eluting with hexane/EtOAc 8:2) gave the corresponding aldol product.

The **diastereomeric ratio** was determined by ¹H NMR of the crude samples after removal of resin catalyst **4** by filtration and evaporation of the solvents.

The **enantiomeric excess** was determined by HPLC on a chiral stationary phase, after flash chromatography on silica gel.

^1H NMR (crude samples, for diastereoselectivity evaluation) and chromatographic data (purified samples, for ee evaluation) of the aldol products:

(2S,1'R)-2-(Hydroxy(phenyl)methyl)cyclohexan-1-one (**s1**):

Chiralcel OD-H column (99:1 Hexane:2-propanol), 1 mL/min; $\lambda = 213$ nm; major enantiomer $t_r = 26.3$ min, minor enantiomer $t_r = 40.2$ min. All the spectroscopic data matched with literature. [S3]

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(2S,1'R)-2-(Hydroxy(naphtyl)methyl)cyclohexan-1-one (**s2**):

Enantiomeric excess was determined by HPLC with a Chiralcel AS column (98:2 Hexane:2-propanol), 1 mL/min; λ = 254 nm; major enantiomer t_r = 32.5 min, minor enantiomer t_r = 39.5 min.

All the spectroscopic data matched with literature. [S3]

(2*S*,1'*R*)-2-(Hydroxy(4-nitrophenyl)methyl)cyclohexan-1-one (**s3**):

Enantiomeric excess was determined by HPLC with a Chiralcel AD-H column (80:20 Hexane:2-propanol), 1 mL/min; $\lambda = 254$ nm; major enantiomer $t_r = 24.1$ min, minor enantiomer $t_r = 31.8$ min.

All the spectroscopic data matched with literature. [S4]

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(2*S*,1'*R*)-2-(Hydroxy(4-methoxyphenyl)methyl)cyclohexan-1-one (**s4**):

Enantiomeric excess was determined by HPLC with a Chiralcel OD-H column (95:5 Hexane:2-propanol), 0.25 mL/min; $\lambda = 254$ nm; major enantiomer $t_r = 77.7$ min, minor enantiomer $t_r = 110.6$ min.

All the spectroscopic data matched with literature. [S5]

(2*S*,1'*R*)-2-(Hydroxy(4-bromophenyl)methyl)cyclohexan-1-one (**s5**):

Enantiomeric excess was determined by HPLC with a Chiralcel AS column (88:12 Hexane:2-propanol), 0.5 mL/min; $\lambda = 254$ nm; major enantiomer $t_r = 13.2$ min, minor enantiomer $t_r = 18.5$ min.

All the spectroscopic data matched with literature. [S6]

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(2*S*,1'*R*)-2-(Hydroxy(-2-furylmethyl)cyclohexan-1-one (**s6**):

Enantiomeric excess was determined by HPLC with a Chiralcel OD-H column (95:5 Hexane:2-propanol), 1 mL/min; $\lambda = 220$ nm; major enantiomer $t_r = 28.9$ min, minor enantiomer $t_r = 30.3$ min.

All the spectroscopic data matched with literature. [S7]

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(2*S*,1'*R*)-2-(Hydroxy(4-trifluoromethylphenyl)methyl)cyclohexan-1-one (**s7**):

Enantiomeric excess was determined by HPLC with a Chiralcel OD-H column (95:5 Hexane:2-propanol), 1 mL/min; λ = 213 nm; major enantiomer t_r = 11.0 min, minor enantiomer t_r = 13.4 min.

All the spectroscopic data matched with literature. [S8]

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(2S,1'R)-2-(Hydroxyphenylmethyl)cyclopentan-1-one (**s8**):

Enantiomeric excess was determined by HPLC with a Chiralcel OD-H column (90:10 Hexane:2-propanol), 1 mL/min; $\lambda = 213$ nm; major enantiomer $t_r = 11.6$ min, minor enantiomer $t_r = 14.0$ min
All the spectroscopic data matched with literature. [S3]

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Recycling experiments of resin 4:

After performing the general procedure for the aldol reaction of cyclohexanone with benzaldehyde, resin **4** was separated by filtration and rinsed with AcOEt (4x2 ml), dried *in vacuo* at 40 °C for 24 h, and reused in the next run. Results have been collected in the following table:

Cycle	Yield (%) ^a	d.r. ^b	ee <i>anti</i> (%) ^c
1	75	96:4	98
2	74	96:4	98
3	75	97:3	98
4	75	97:3	98
5	75	97:3	98

^a Isolated Yield. ^b Determined by ¹H NMR. ^c Determined by chiral HPLC.

Aldol reaction at different temperatures using resin 4:

The aldol reaction of cyclohexanone and benzaldehyde mediated by resin **4** was performed at different temperatures following the general procedure described above. Results have been collected in the following Table:

entry	T (°C)	Time (h)	Yield (%) ^a	d.r. ^b	ee <i>anti</i> (%) ^c
1	25	24	74	96:4	98
2	40	12	70	93:7	97
3	60	6	55	85:15	92

^a Isolated Yield. ^b Determined by ¹H NMR. ^c Determined by chiral HPLC.

Procedure for the self-aldol addition of propanal in water catalyzed by resin

4.[S9]

Propionaldehyde (0.6 mL, 8 mmol) was added to resin **4** (71 mg, 1 mol%, $f=0.56$ mmol/g) in a vial with 2.87 mL of water (20 equiv.). The mixture was stirred at r.t. for 24 h. A sample was analyzed by ^1H -NMR, and showed total conversion (no propionaldehyde could be detected). Then the resin was removed by filtration, and rinsed with CH_2Cl_2 . The solution containing the crude product was treated with a cooled suspension of NaBH_4 (0.45 g, 12 mmol) in EtOH (20 mL) at 0°C . After 1 hour the reaction was diluted with water (20 mL) and extracted with CH_2Cl_2 (3 x 20 mL). After drying with Na_2SO_4 , solvents were removed *in vacuo*. ^1H -NMR of the crude product showed a diastereomeric ratio of diols of anti:syn 4:1. After purification by flash chromatography on silica gel, 100 mg of the diol were isolated as a mixture of diastereomers (21% yield). The spectroscopic data matched with the literature data.[S9] (Note that dehydration products were formed during the chromatographic process).

The diol was derivatized as monobenzoyl ester, purified by flash chromatography on silica gel, and analyzed by HPLC (Chiralpak IA, hexane:IPA 99:1, 1 mL/min, 230 nm): t_R = 29.4 and 36.3 min (*syn*), and 39.8 and 42.8 min (*anti*, 97% ee).

a) Racemic **s9** for reference purposes

b) Enantioenriched **s9** from the reaction catalyzed by **4**.

Atomic Coordinates for Proline-triazole model with two water molecules

Computed with Gaussian 03 Revision-C.02, with an ONIOM method at the MP2 level for the inner set of atoms involving hydrogen bonding, and at the PM3 level for the outer atoms (ONIOM (MP2-FC/6-31G(d,p):PM3/ZDO)):

Most stable conformer (E = -677.68809314 a.u., number of imaginary frequencies=0)

Center Number	Atomic Number	Coordinates (Angstroms)		
		X	Y	Z
1	6	-2.943895	1.928499	-2.455909
2	6	-3.928997	0.234537	-0.472333
3	6	-3.278167	0.614008	-2.765698
4	6	-3.102714	2.396764	-1.156120
5	6	-3.593676	1.553052	-0.165321
6	6	-3.771226	-0.231209	-1.778337
7	1	-3.161113	0.246406	-3.790399
8	1	-2.843098	3.432092	-0.911378
9	1	-3.718436	1.927063	0.857649
10	1	-4.041022	-1.265281	-2.021430
11	6	-4.493376	-0.670856	0.586185
12	1	-4.731380	-0.107774	1.510613
13	1	-5.420964	-1.151697	0.226198
14	8	-3.685484	-1.800181	0.864005
15	6	-2.654433	-1.616676	1.814263
16	1	-2.982959	-0.974821	2.656310
17	1	-2.519690	-2.645922	2.196292
18	6	-1.383528	-1.128715	1.222903
19	6	-0.773872	-1.513433	0.042821
20	1	-1.058781	-2.196290	-0.738167
21	7	0.364066	-0.773308	0.010597
22	7	0.489046	0.021176	1.102162
23	7	-0.591669	-0.200752	1.843024
24	6	1.427474	-0.816385	-1.038299
25	6	1.881883	0.601370	-1.456131
26	7	3.026741	0.942816	-0.561946
27	6	3.581844	-0.325033	-0.004090
28	6	2.719853	-1.476971	-0.502100
29	1	1.004253	-1.382987	-1.900950
30	1	1.085580	1.367705	-1.413717
31	1	2.254182	0.585806	-2.499055
32	1	3.571071	-0.279896	1.092546
33	1	2.532083	-2.227959	0.288059
34	1	3.213596	-2.035799	-1.322077
35	1	2.725033	1.537848	0.219296
36	6	5.045039	-0.511644	-0.454494
37	8	5.721447	-1.469331	-0.128070
38	8	5.489194	0.495771	-1.220654
39	1	4.702502	1.094213	-1.287202
40	1	3.413920	2.411982	2.563986
41	8	2.657529	2.317020	1.978231
42	1	1.935733	2.006092	2.557084
43	1	-0.020743	2.454140	3.616401

44	8	0.530158	1.673035	3.732248
45	1	0.025292	0.965528	3.292664
46	1	-2.561787	2.595305	-3.235799

Less stable conformer (E = -677.67570510 a.u., number of imaginary frequencies=0)

Center Number	Atomic Number	Coordinates (Angstroms)		
		X	Y	Z
1	6	-3.756162	-0.839196	0.303208
2	7	-2.685905	-1.729587	0.825336
3	6	-1.718945	-1.943843	-0.285441
4	6	-1.780148	-0.647184	-1.134579
5	6	-3.236858	-0.158265	-0.968832
6	1	-3.996931	-0.084951	1.059607
7	1	-0.710971	-2.199223	0.089987
8	1	-2.065556	-2.807557	-0.886240
9	1	-1.537729	-0.849957	-2.204380
10	1	-3.823941	-0.450663	-1.862141
11	1	-3.330108	0.944492	-0.910756
12	1	-2.185766	-1.230751	1.554312
13	6	-5.035869	-1.623397	-0.041132
14	8	-5.055075	-2.818794	-0.252280
15	8	-6.142420	-0.860598	-0.136267
16	7	-0.782098	0.359499	-0.658338
17	6	0.415939	0.635689	-1.233793
18	1	0.746830	0.166024	-2.143313
19	6	0.980599	1.577543	-0.390672
20	7	0.120529	1.828500	0.640957
21	7	-0.959980	1.074608	0.482431
22	6	2.294191	2.262636	-0.516527
23	1	2.359219	3.153056	0.135220
24	1	2.449059	2.583915	-1.565971
25	8	3.378130	1.493859	-0.042394
26	6	3.809429	0.465821	-0.928849
27	1	2.969577	-0.220718	-1.158435
28	1	4.143184	0.911861	-1.887332
29	6	4.926283	-0.266870	-0.245343
30	6	6.999370	-1.642854	1.009013
31	6	4.642061	-1.232623	0.720566
32	6	6.252222	0.010517	-0.577861
33	6	7.285100	-0.678835	0.048047
34	6	5.677933	-1.917811	1.345491
35	1	3.601694	-1.444618	0.990641
36	1	6.480925	0.774401	-1.329641
37	1	8.325018	-0.459222	-0.214783
38	1	5.452159	-2.673187	2.105223
39	1	7.813960	-2.183355	1.501937
40	8	-5.712715	1.780304	0.428203
41	8	-3.317544	2.337623	1.706678
42	1	-3.135234	2.294335	2.650068
43	1	-4.895900	2.045328	0.895519
44	1	-2.540621	1.931293	1.282832
45	1	-6.435977	2.169577	0.928453
46	1	-5.948887	0.084206	0.068110

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